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TITLE: Quantitative, Noninvasive Imaging of DNA Damage *in Vivo* of Prostate Cancer Therapy by Transurethral Photoacoustic (TUPA) Imaging

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14. ABSTRACT **The goals of this training grant is to develop the foundations for a new medical imaging modality, now called Trans-Urethral Photoacoustic (TUPA) Imaging, which utilizing a small catheter into the urethra enabling imaging molecular marker of DNA Damage during prostate radiation therapy with high photoacoustic contrast and resolution. The development of this modality is significant to tracking the biologic effect of radiation treatment, rather than just the physical dose1-6. The first-year research goals were to develop the imaging instrumentation to enable this modality. This grant has provided the funding to devise a trans-urethral photoacoustic endoscope, which has the potential to obtain higher resolution by using a high frequency ultrasound detector and achieve deeper penetration depth with a light delivery via the urethra. For the first-year training goals, this grant has provided for extensive study in prostate cancer, molecular imaging modalities, molecular targeting of cancer, and opportunities to engage physicians. After finished the 1st year project at Stanford University (30 SEP 2013 -31 DEC 2014). I have now accepted a tenure-track assistant professor position at The University of Oklahoma officially start from February in 2015. More than 1000 square feet of new lab space was assigned to me at Stephenson Research & Technology Center. I want to finish the DoD prostate cancer project and continue to do research at the new institution. The preparations have been done for transferring this grant from Stanford to The University of Oklahoma: 1000+ square feet of lab space is opening at The University of Oklahoma; 1 postdoctoral research fellow was hired; collaboration with Urologist at OUHSC has been build, and the photoacoustic (TUPA) Imaging system has been rebuilt in The University of Oklahoma. As of today, the results of this grant are: 2 peer reviewed journal publications, and 1 paper accepted; 5 conference abstracts, including 3 first author, 1 corresponding author, and 2 conference oral presentations. The future for this grant looks bright, as this technique will soon be tested in small animal models in vivo; the next, more clinically feasible interventional version is under development.**

15. SUBJECT TERMS

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1. INTRODUCTION

The goals of this training grant is to develop the foundations for a new medical imaging modality, now called Trans-Urethral Photoacoustic (TUPA) Imaging, which utilizing a small catheter into the urethra enabling imaging molecular marker of DNA Damage during prostate radiation therapy with high photoacoustic contrast and resolution. The development of this modality is significant to tracking the biologic effect of radiation treatment, rather than just the physical dose[1-6].

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2. KEYWORDS

Prostate Cancer, Photoacoustic Imaging, Molecular Imaging, Radiation Therapy

3. OVERALL PROJECT SUMMARY

Research Accomplishments: SOW Aim 1: Develop a photoacoustic imaging system for prostate cancer imaging

Major Task 1: Develop photoacoustic imaging system

Subtask 1: Investigation of photoacoustic imaging system (month 1-6)

The trans-urethral photoacoustic (TUPA) catheter has been developed in the first 6 month of this project, shown in Figure 1. Figure 1(a) is a schematic to illustrate the principle of the mechanical scanning and the configuration of the optical and acoustic components. The catheter was built with a side-fire optical fiber for light delivery and a commercially available mini ultrasound transducer. The catheter was capable of both receiving the photoacoustic signal and performing ultrasound pulse-echo imaging[8].

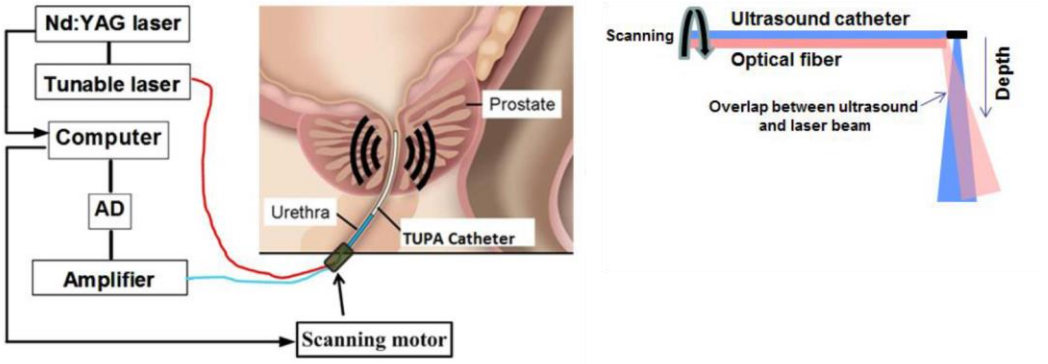


Figure 1 (a) Schematic of trans-urethral photoacoustic (TUPA) system

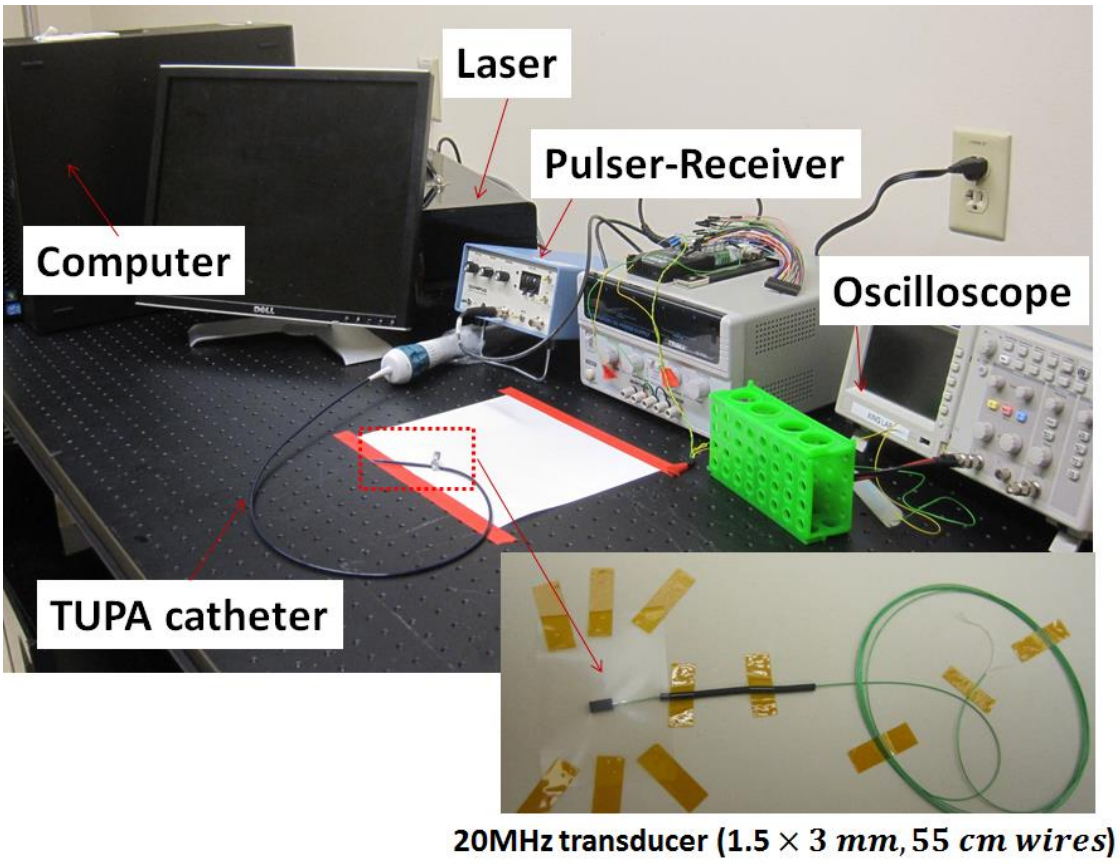


Figure 1 (b) Picture of the trans-urethral photoacoustic (TUPA) system and its key components

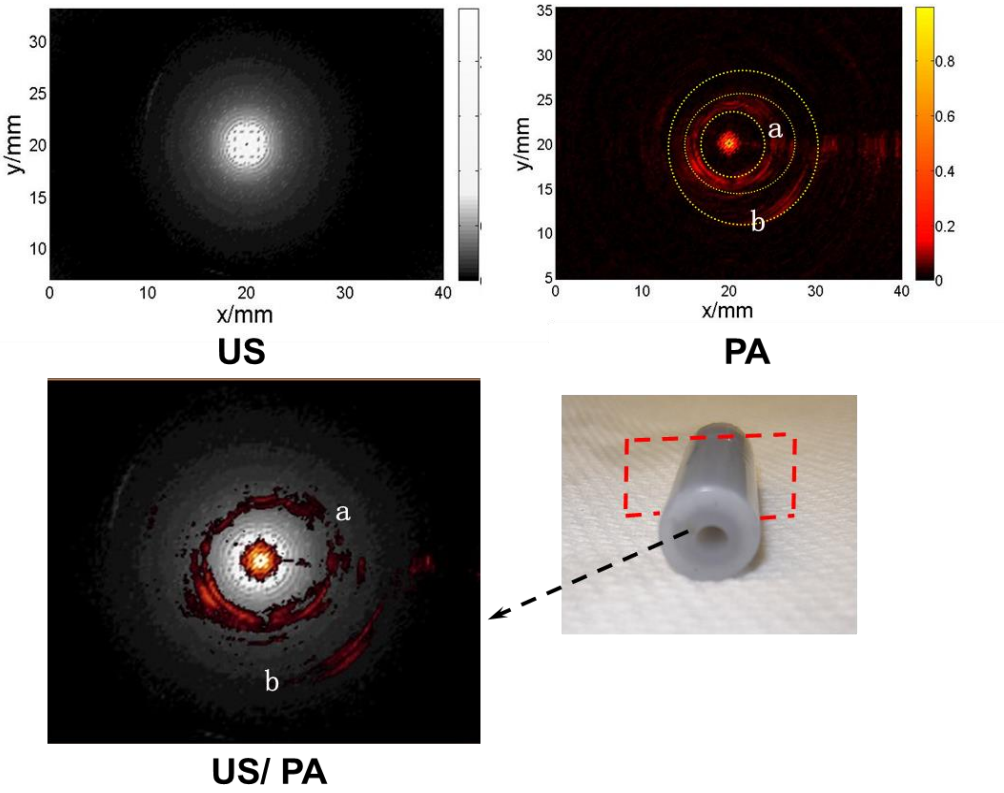


Figure 1 (c) Phantom experiments by using trans-urethral photoacoustic (TUPA) system. US : ultrasound imaging of the phantom; PA: Photoacoustic imaging of the phantom; and combined US/PA imaging of the phantom; and the picture of the phantom.

Figure 1(b) shows photos of trans-urethral photoacoustic (TUPA) imaging system. A TUPA catheter comprised of two key components: optical laser fiber, and the mini ultrasound transducer. An ultrasonic (US) transducer (LiNbO3, ~20 MHz, unfocused) generates US pulses and detects both PA and US pulse-echo signals. The US transducer was costumed made by Blatek, Inc. with a very small piezoelement size (1.5x3 mm). It provides two 55cm wires which can send the photoacoustic signal outside of the catheter to the ultrasound pulser and receiver (5072PR, Olympus). Inside the tubular shaft, a multimode optical fiber (UM22-600, Thorlabs) was placed which is positioned statically along the axis of the endoscope. A parabolic acoustic reflector (10 mm diameter, nickel substrate, Optiforms) and an optical prism (3 mm diameter, altered from #45-525, Edmund) was mounted inside the catheter. The scanning head is actuated by a step motor located at the proximal end. Via the prism and parabolic reflector, laser pulses and acoustic waves are delivered coaxially to achieve an efficient overlap of the illumination and acoustic detection over a large depth range. A membrane forms an imaging window and seals the inner cavity of the endoscope, which is filled with de-ionized water. The imaging capability of combined PA and US imaging with endoscopic catheter has been demonstrated on a phantom imbedded with IRDye 800CW as a contrast agent in the sample (Figure 1(c)).

This work was carried out under guidance from my mentor Dr. Lei Xing, as proposed, who is an expert in medical imaging instrumentation.

Subtask 2: Control software will be developed in Labview to enable automated data acquisition (month 7-12)

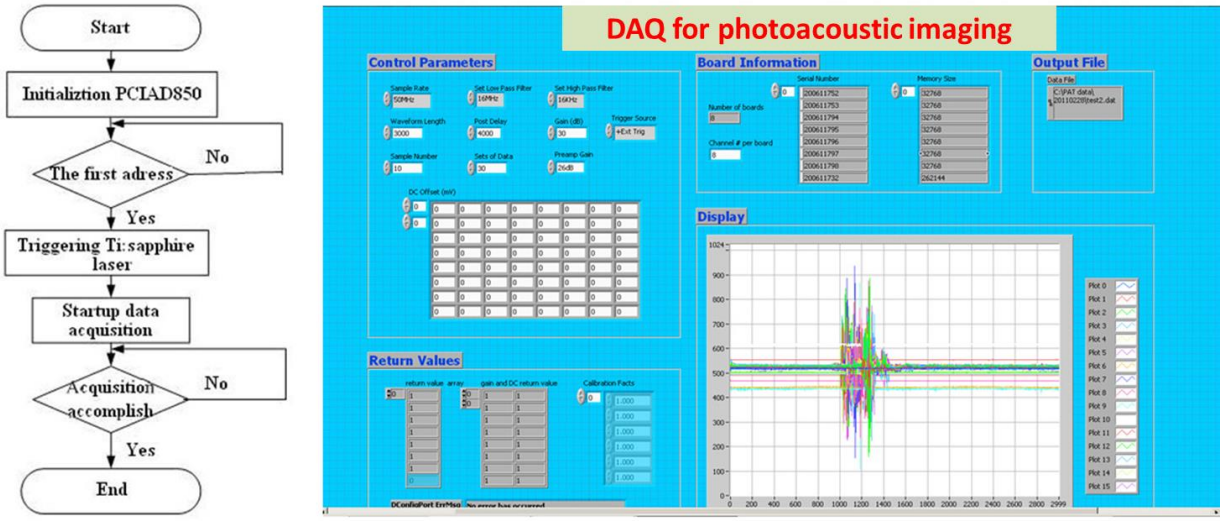


Figure 2 Flowchart of the photoacoustic data acquisition loop, and control panel of data acquisition software for photoacoustic imaging

A Labview based platform was built to control the data acquisition system (Figure 2). The 2-D PA/US image was reconstructed with the modified filtered back projection algorithm constructed in the MATLAB programming language. Light and ultrasound attenuation was considered during the imaging reconstruction. Image registration between PA and US has been done automatically in the software. The reconstructed data from a pullback scan can then be used to generate a 3-D PA/US image.

Training Accomplishments:

Major Task 1: Training and educational development in prostate cancer research

Subtask 1: Attend radiation physics resident lecture series *(month 1-6)*

Stanford University Radiation Oncology has a comprehensive training program that emphasizes on research with ambitious clinical goals. I attended weekly research seminars (every Tuesday morning) held by the department, where Stanford faculty members, postdoctoral fellows, as well as invited speakers from outside give presentations and open discussions on their research. Some of the seminars are very impressive to me. For example, one of the seminars titled as “Image Guidance in Radiation Therapy: Applications to Adaptive Treatment Planning and Response” is really help me understand the current clinical needs in radiation therapy. In addition, to keep up with the fast development of radiation physics research, I also attended biweekly radiation physics journal club meetings to discuss and exchange ideas about the most recent research. The comprehensive training program and an easily accessible talent pool at Stanford have contributed significantly to the success of the proposed research.

Subtask 2: Present research at the monthly department group meetings *(month 1-12)*

It is required to present my research at the monthly department group meetings here at Stanford. My mentor Dr. Lei Xing and other members have been invited. In those group meetings, we discussed the progress of current projects and the latest progresses in prostate cancer research, molecular imaging, bioengineering, or related fields. After each presentation, the advisory team provided constructive feedback and advice on his research and presentation skills. I was also invited to present my research at the Stanford Radiation Biology Seminar on August 12th, 2014 titled as "Radiation induced acoustic emission for cancer imaging and diagnosis".

Subtask 3: Attend a national scientific meeting in relevant scientific field *(month 1-12)*

I attended the annual meetings held by professional societies in the field, such as American Society of Therapeutic Radiology and Oncology (ASTRO) on 09/15/2014 at San Francisco, California, and American Association of Physicists in Medicine (AAPM) on 07/22/2014 at Texas Austin, etc. These scientific meetings offer an ideal platform for me to communicate and exchange ideas about the latest development related to the proposed research with peers around the globe.

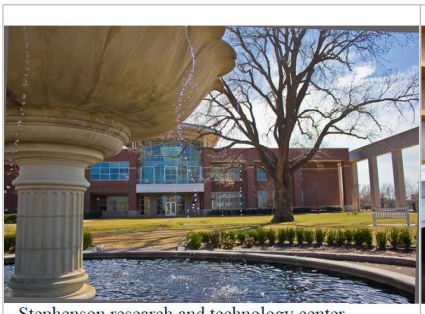
In performing these tasks, the investigator has been exposed to the field of molecular imaging, a new direction for this PI. This research education has been aided with participation in BioE222: Molecular Imaging, which brought together the top molecular imaging faculty at Stanford to teach aspects in the hardware, chemistry, and biology of molecular imaging. In addition, the PI was exposed to molecular probe fabrication, including the processes in making IRDye stable in human serum with low toxicity.

Also, the PI gained knowledge in molecular targets, and the advantages and disadvantages of targeting to peptides, hormones, antibidies, affabodies, and other targeting agents. This program has been aided by working at benchside with these materials scientists, biologists, and molecular imaging experts.

Preparation for transferring the grant:

Subtask 1: 1000+ square feet of lab space is opening at The University of Oklahoma

More than 1000 square feet of lab space is opening for continuing this project in Stephenson research and technology center at University of Oklahoma. This significant step will enable building up our TUPA imaging systems as well as prepare the TRUE lab for future growth. See details in our lab website: https://sites.google.com/site/xianglzh/news/1000squarefeetoflabspaceisopening



Stephenson research and technology center

Electronics&. Mechanics bench

Biological sample &. Phantom preparation bench

Interior view of the Stephenson research and technology center | Chemicals preparation bench | Equipment storage

Subtask 2: Dr. Shanshan Tang as a postdoc has been hired for the project with expertise in medical ultrasound imaging technique

The primary role of the postdoc is to rebuild and optimize transurethral photoacoustic (PA) and ultrasound (US) endoscopy at The University of Oklahoma. She will establish the necessary supporting peripheral subsystems including a laser source and light delivery path, a data acquisition subsystem, and a master control of all subsystems. She will be under the direct supervision of Dr. Xiang, and work closely with our collaborators, Drs. Stratton and Cookson in the Urology Department at OUHSC. The post-doc will support this project with 8.0 person months in the year (66.7%).

Subtask 3: Urologist Drs. Michael Cookson and Kelly Stratton as collaborators in the OUHSC will help performing the project in the animal experiments and the future clinical investigations.

As Professor and Chair in Department of Urology at University of Oklahoma College of Medicine, and Director of Urologic Oncology at the Stephenson Cancer Center, Dr. Michael Cookson's group is working on the prostate cancer detection and treatment. He will share his clinical research experience to serve in the role of collaborator in this project. Dr. Kelly Stratton, as an assistant professor will primarily devoted to working in this collaboration (support letter are attached). This collaboration is critical for finishing the aim 3 for this project.

4. KEY RESEARCH ACCOMPLISHMENTS

- Developed a trans-urethral photoacoustic (TUPA) imaging, in an appropriate geometry for monitoring of radiation therapy for prostate cancer patient.
- Fabricated a trans-urethral photoacoustic (TUPA) imaging catheter to enable automated, controlled imaging of the DNA damage during prostate cancer radiation therapy.
- A Labview based software platform was built to control the data acquisition system for photoacoustic imaging.

5. CONCLUSION

The first funding period in this grant has developed the research infrastructure for trans-urethral photoacoustic (TUPA) imaging. This has resulted in a fully functioning system that may perform the systematic studies in phantoms and pre-clinical animals that is the crux of Aims 3 from the SOW. Towards the goal of determining the feasibility for patient

imaging, and we will provide proof-of-concept for the prostate cancer treatment monitoring realizations of this modality. Future work for Aims 3 will involve a more sophisticated tissue-simulating phantom study to evaluate system temporal and sensitivity performance with regards to imperfect background contrast uptake, and a systematic study to determine the feasibility in pre-clinical and clinical research. In addition, the first funding period for this grant resulted in much training for the PI, for molecular imaging, small-animal imaging, cancer biology, and device commercialization. The research and training in the grant are significant for the eradication of prostate cancer for the new developments in the potential for early detection of prostate cancer, for the treatment of prostate cancer by providing feedback to guide radiation therapy, and for the coursework which will enable this PI to apply newly learned skills to bring technologies to the clinic and marketplace. It is our hope that this pioneering work will lead to advancements in this new field that can be translated to the clinic.

6. PUBLICATIONS, ABSTRACTS, AND PRESENTATIONS

2 peer reviewed journal publications, and **1** paper accepted; **5** conference abstracts, including **3** first author, **1** corresponding author, and **2** conference oral presentations; 4 courses taken, including BioE222: Molecular Imaging, Med374: Medical Device Design. The future for this grant looks bright, as this technique will soon be tested in small animal models *in vivo*; the next, more clinically feasible interventional version is under development.

Presentations:

- Shanshan Tang, Hong Liu, **Liangzhong Xiang**, Transurethral photoacoustic endoscopy: making moves into the clinic, 2015 World Molecular Imaging Congress, September 2, 2015. Oral Presentation
- **Liangzhong Xiang**, Benjamin P Fahimian, Moiz Ahmad, and Lei Xing, "High precision focal brachytherapy of prostate cancer guided by dual-model photoacoustic and ultrasound imaging" Presented at 56th Annual Meeting, San Francisco, California, on 09/13/2014. Poster
- **L Xiang**, M Ahmad, A Nikoozadeh, G Pratx, B Khuri-Yakub, L Xing, "X-ray acoustic computed tomography (XACT): 100% sensitivity to X-ray absorption", Presented at 56th Annual Meeting&. Exhibition, Austin, Texas, on 07/22/2014. Oral Presentation
- **Liangzhong Xiang**, M Ahmad, C Carpenter, G Pratx, A Nikoozadeh, B Khuri-Yakub, L Xing, "X-Ray Acoustic Computed Tomography: Concept and Design", Presented at 55th Annual Meeting&. Exhibition, Indianapolis, Indiana, on 08/08/2013. Hot Topic Oral Presentation
- M Ahmad, M Bazalova, **L Xiang**, L Xing, "X-Ray Fluorescence CT as a Novel Imaging Modality for Improved Radiation Therapy Target Delineation", Presented at 56th Annual Meeting, San Francisco, California, on 09/13/2014. Poster

Journal Publications:

- Junping Zhong, Liewei Wen, Sihua Yang**, Liangzhong Xiang**, Qun Chen, Imaging-guided high-efficient photoacoustic tumor therapy with targeting gold nanorods, Journal of Nanomedicine: Nanotechnology, Biology, and Medicine, Accepted, 2015.
- **Liangzhong Xiang**, Moiz Ahmad, Xiang Hu, Zhen Cheng, and Lei Xing, Label-free photoacoustic cell-tracking in real-time, *X-Acoustics: Imaging and Sensing*, 1: 18-22 (2014).
- Moiz Ahmad, Magdalena Bazalova, **Liangzhong Xiang**, and Lei Xing, Order of magnitude sensitivity increase in x-ray fluorescence computed tomography (XFCT) imaging with an optimized spectro-spatial detector configuration: theory and simulation, *IEEE Trans. Med. Imag.*, 99, (2014).

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7. Liangzhong Xiang, Moiz Ahmad, Xiang Hu, Zhen Cheng, and Lei Xing, Label-free photoacoustic cell-tracking in real-time, *X-Acoustics: Imaging and Sensing*, 1: 18-22 (2014).
8. Liangzhong. Xiang, Benjamin P Fahimian, Moiz Ahmad, Mark Buyyounouski, James Brooks, Lei. Xing, Photoacoustic and ultrasound image-guided focal brachytherapy of prostate cancer, in preparation, (2014).

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Chairman and Professor
Donald D. Albers Endowed Chair
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Urologic Oncology

March 18, 2015

Liangzhong Xiang, Ph.D.
School of Electric and Computer Engineering
University of Oklahoma
101 David L Boren Blvd, Room 2022
Norman, OK 73019, USA
Office phone: (405) 325-7148

Dear Liangzhong:

It is with great pleasure that I write this letter to support your DoD project, “Quantitative, Noninvasive Imaging of DNA Damage *in Vivo* of Prostate Cancer Therapy by Transurethral Photoacoustic (TUPA) Imaging”. As Professor and Chair in Department of Urology at University of Oklahoma College of Medicine, and Director of Urologic Oncology at the Stephenson Cancer Center, my group is working on the prostate cancer detection and treatment. I am happy to share my clinical research experience to serve in the role of collaborator in this project.

I am very enthusiastic about the photoacoustic and ultrasound endoscopy approach that you are proposing. This approach promises a new paradigm for the detection of prostate cancer. I am therefore particularly confident that photoacoustic and ultrasound endoscopy can help the prostate therapy monitoring. It will significantly improve clinical practice of prostate cancer management and deliver tangible benefits to patients. I also believe this transurethral photoacoustic and ultrasound endoscope is directly translatable to clinical practice.

My group will provide you expertise in the management of prostate cancer to achieve Specific Aims 3 of your project. Specifically, I will arrange for you to observe actual prostate detection and treatment procedures in clinic. I will also provide my help in making the prostate phantom and provide the help for the animal experiments. Dr. Kelly Stratton, as an assistant professor will primarily devoted to working in this collaboration. Please do not hesitate to contact me any time. I look forward to a fruitful collaboration in this exciting project.

Sincerely,

Cookson S. Michael, MD, MMHC
Professor, Department of Urology
Donald D. Albers, MD Chair in Urology